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Research Note

Endoparasites of the Red-backed Salamander, *Plethodon c. cinereus*, from Southwestern Michigan

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ABSTRACT: Three species of endoparasites were found in 171 red-backed salamanders, *Plethodon c. cinereus*, collected from southwestern lower Michigan between 27 March and 2 September 1989. The nematode, *Thelandros magnavulvaris*, had the highest prevalence (28%), and the trematode, *Brachycoelium salamandrae*, had the highest mean intensity (2.9). The ciliate, *Cepedietta michiganensis*, infected 18% of the salamanders. Michigan is a new locality record for *T. magnavulvaris*.

KEY WORDS: *Plethodon c. cinereus*, red-backed salamander, endoparasites, survey, Nematoda, Protozoa, Trematoda, Michigan.

Although the parasites of the red-backed salamander, *Plethodon c. cinereus* (Green) have been studied by several authors, most notably Rankin (1937a, b, 1945), Walton (1938), and Fischthal (1955a, b), little is known about the parasites of this terrestrial salamander from the Great Lakes area. This note presents new information on the parasites of the red-backed salamander from this region and increases the information known about the parasites of Michigan salamanders.

One hundred seventy-one red-backed salamanders were collected by hand from the Barry Game Area, Barry County, southwestern lower Michigan, between 27 March and 2 September 1989. Both color phases (red-backed and lead-backed) of this salamander species were collected in a mature forest of beech, maple, and oak trees east of Otis Lake. Salamanders were killed in MS222 (ethyl m-aminobenzoate methane sulfonic acid). The head–body length (mm), color phase, and sex were recorded before the entire salamander was necropsied within 18 hr of collection. The mean head–body length ±1 SD (range) of all red-backed salamanders examined was 40 ± 6.8 mm (20–55 mm). Parasites found were processed using conventional parasitologic techniques. Prevalence is the percentage of animals infected in a sample and mean intensity is the mean number of worms per host. Representative specimens of parasites from salamanders have been deposited in the U.S. National Parasite Collection, Beltsville, Maryland (accession nos. 80995–80997).

Ninety-one (53%) red-backed salamanders were infected with 1 or more *Thelandros magnavulvaris* (Rankin, 1937), *Brachycoelium salamandrae* (Frölich, 1789), and *Cepedietta michiganensis* Woodhead, 1928. *Thelandros magnavulvaris* had the highest prevalence, and *B. salamandrae* had the highest mean intensity (Table 1). Nine hosts (10%) were concurrently infected with *T. magnavulvaris* and *B. salamandrae*, 5 (5%) with both *T. magnavulvaris* and *C. michiganensis*, and 1 (1%) with both *B. salamandrae* and *C. michiganensis*. Although at least 20 salamanders were collected each month, infection values for each parasite species were low and/or erratic over the 7-mo period. The prevalence and mean intensity of *T. magnavulvaris* were highest in April. The prevalence and mean intensity of *B. salamandrae* were highest in September and July, respectively. The prevalence of *C. michiganensis* was highest in August. There were no significant differences in prevalence and intensity of parasitism between females and males, nor between the 2 color phases (chi-square analysis and Student's *t*-test). There were also no distinct increases in infection for each parasite species with salamander length.

Thelandros magnavulvaris (=*Batracholandros magnavulvaris* as indicated by Petter and Quentin [1976]) has been reported from a variety of salamanders by Rankin (1937a, b), Lehmann (1954), Schad (1963), Fischthal (1955a, b), Dyer and Peck (1975), Dunbar and Moore (1979), and Dyer et al. (1980). Michigan is a new locality record for *T. magnavulvaris* and extends its range northward. Dunbar and Moore (1979) reported that red-backed salamanders and other terrestrial species were not infected with *T. magna-*

Table 1. Prevalence and mean intensity of parasites found in 171 *Plethodon c. cinereus* from the Barry Game Area.

Parasite	Prevalence	Mean intensity ±1 SD (range)	Site of infection	Mean length (mm) ± 1 SD (range) of infected *P. c. cinereus*
Thelandros magnavulvaris	48 (28)*	1.9 ± 1.3 (1–7)	cloaca	43 ± 5.6 (31–55)
Brachycoelium salamandrae	26 (15)	2.9 ± 3.3 (1–16)	small intestine	41.9 ± 6.3 (33–54)
Cepedietta michiganensis	31 (18)	—	small intestine, gall bladder	42.1 ± 5.4 (29–53)

* Number infected (percent infected).

vulvaris, whereas the aquatic to semiaquatic and semiterrestrial salamanders were infected. As true in other studies on *Thelandros* spp., female *T. magnavulvaris* were much more common than males in red-backed salamanders.

Although the trematodes collected in the present study exhibited much morphological variation, they were identified as *B. salamandrae* using the information presented by Byrd (1937) and the key of Cheng (1958). In Michigan, *B. salamandrae* has been found in the salamander *Hemidactylium scutatum* by Rankin (1938) and in the frogs *Acris gryllus* and *Rana sylvatica* by Najarian (1955). Coggins and Sajdak (1982) reported *B. salamandrae* in the marbled salamander, *Ambystoma opacum,* and in red-backed salamanders from Wisconsin.

Cepedietta michiganensis (Haptophryidae) was originally described from *H. scutatum* from southeastern Michigan by Woodhead (1928). Blanchard (1923) found approximately 70% of several thousand *H. scutatum* and 1 *Ambystoma jeffersonianum* from Michigan infected with this ciliate. Since then, *C. michiganensis* has been found in other plethodontid salamanders by Hazard (1937), Rankin (1937a, b), and Powders (1967, 1970) and in *R. sylvatica* by Hazard (1937). Woodhead and Kruidenier (1936) reported that larval *H. scutatum* ingested the active protozoans in fecal matter and carried them through metamorphosis to the adult stage. Hazard (1937) suggested that red-backed salamanders became infected in the same way. In the present study, all infections of red-backed salamanders by *C. michiganensis* were very heavy, with hundreds of protozoans found. Ciliates in the gall bladder were easily observed with the dissecting microscope.

The results of the present study are similar to those of other parasitologic surveys of *P. c. cinereus* by Rankin (1945), Fischthal (1955a, b), Dunbar and Moore (1979), and Coggins and Sajdak (1982) in that the number of parasite species found is low and the number of red-backed salamanders concurrently infected with 2 or more parasite species is low. The most parasite species found in a population of red-backed salamanders was by Rankin (1937a) who reported 9 protozoans and 4 helminths.

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Literature Cited

Blanchard, F. N. 1923. The life history of the four-toed salamander. American Naturalist 57:262–268.

Byrd, E. E. 1937. Observations on the trematode genus *Brachycoelium* Dujardin. Proceedings of the United States National Museum 84:183–199.

Cheng, T. C. 1958. Studies on the trematode family Dicrocoeliidae. I. The genera *Brachycoelium* (Dujardin, 1845) and *Leptophallus* Luhe, 1909, (Brachycoeliinae). American Midland Naturalist 59:67–81.

Coggins, J. R., and R. A. Sajdak. 1982. A survey of helminth parasites in the salamanders and certain anurans from Wisconsin. Proceedings of the Helminthological Society of Washington 49:99–102.

Dunbar, J. R., and J. D. Moore. 1979. Correlations of host specificity with host habitat in helminths parasitizing the plethodontids of Washington County, Tennessee. Journal of the Tennessee Academy of Science 54:106–109.

Dyer, W. G., R. A. Brandon, and R. L. Price. 1980. Gastrointestinal helminths in relation to sex and age of *Desmognathus fuscus* (Green, 1818) from Illinois. Proceedings of the Helminthological Society of Washington 47:95–99.

———, **and S. B. Peck.** 1975. Gastrointestinal parasites of the cave salamander, *Eurycea lucifuga* Rafinesque, from the southeastern United States. Canadian Journal of Zoology 53:52–54.

Fischthal, J. H. 1955a. Helminths of salamanders from Promised Land State Forest Park, Pennsyl-

vania. Proceedings of the Helminthological Society of Washington 22:46–48.

———. 1955b. Ecology of worm parasites in south-central New York salamanders. American Midland Naturalist 53:176–183.

Hazard, F. O. 1937. Two new host records for the protozoan *Haptophrya michiganensis* Woodhead. Journal of Parasitology 23:315–316.

Lehmann, D. L. 1954. Some helminths of West Coast urodeles. Journal of Parasitology 40:231.

Najarian, H. H. 1955. Trematodes parasitic in the Salientia in the vicinity of Ann Arbor, Michigan. American Midland Naturalist 53:195–197.

Petter, A. J., and J. C. Quentin. 1976. Keys to Genera of the Oxyuroidea. *In* R. C. Anderson, A. G. Chabaud, and S. Willmott, eds. CIH Keys to the Nematode Parasites of Vertebrates. No. 4. Commonwealth Agricultural Bureaux, Farnham Royal, England. 30 pp.

Powders, V. N. 1967. Altitudinal distribution of the astomatous ciliate *Cepedietta michiganensis* (Woodhead) in a new host, *Plethodon jordani* Blatchley. Transactions of the American Microscopical Society 86:336–338.

———. 1970. Altitudinal distribution of the protozoan *Cepedietta michiganensis* in the salamanders *Plethodon glutinosus* and *Plethodon jordani* in eastern Tennessee. American Midland Naturalist 83:393–403.

Rankin, J. S., Jr. 1937a. An ecological study of parasites of some North Carolina salamanders. Ecological Monographs 7:169–270.

———. 1937b. New helminths from North Carolina salamanders. Journal of Parasitology 23:29–42.

———. 1938. Studies on the trematode genus *Brachycoelium* Duj. I. Variation in specific characters with reference to the validity of the described species. Transactions of the American Microscopical Society 57:358–375.

———. 1945. An ecological study of the helminth parasites of amphibians and reptiles of western Massachusetts and vicinity. Journal of Parasitology 31:142–150.

Schad, G. A. 1963. *Thelandros magnavulvaris* (Rankin, 1937) Schad, 1960 (Nematoda: Oxyuroidea) from the green salamander, *Aneides aeneus*. Canadian Journal of Zoology 41:943–946.

Walton, A. C. 1938. The Nematoda as parasites of Amphibia. IV. Transactions of the American Microscopical Society 57:38–53.

Woodhead, A. E. 1928. *Haptophyra michiganensis* sp. nov., a protozoan parasite of the four-toed salamander. Journal of Parasitology 14:177–182.

———, **and F. Kruidenier.** 1936. The probable method of infection of the four-toed salamander with the protozoon, *Haptophrya michiganensis*. Journal of Parasitology 22:107–108.

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Research Note

An Apparatus for Modified Harada-Mori Cultures of Third-stage Hookworm Larvae

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ABSTRACT: An apparatus is described that allows for the filter paper culture of third-stage hookworm larvae, suitable for biochemical studies. The method used consumes less time and space than conventional Harada-Mori test tube cultures, allowing for the application of a large volume of feces over a surface area of up to 6,400 cm² within a box small enough for a bench-top incubator.

KEY WORDS: *Ancylostoma caninum, Necator americanus,* hookworm, nematode larva.

Although large numbers of nematode larvae can be obtained via charcoal culture, they are often contaminated with organic debris and are therefore potentially unsuitable for biochemical studies. To circumvent the problem of contamination, investigators have attempted to separate larvae from fecal sediment by either centrifugation through ficoll-sodium metrizoate (Damian, 1976) or via filter paper cultures in petri dishes (Cross and Scott, 1961; Burren, 1980; Mueller et al., 1989). In the 1950's, Harada and Mori described a method whereby hookworm larvae migrate down filter paper placed in a con-